

AMENDMENTS TO THE CLAIMS

1. (currently amended): A method for stable transduction of a primary lymphoid cell, a myeloid cell or a hematopoietic progenitor cell comprising

contacting the cell surface of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell at the same time *in vitro* or *ex vivo* with both a lentiviral vector and at least one cell stimulatory polypeptide which binds the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell by binding to a cell surface protein or to a cell surface receptor,

wherein the at least one cell stimulatory polypeptide is an antibody, an antigen binding fragment, or a ligand, and

wherein after the contacting, at least ~~about 75%~~ **90%** of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cells are stably transduced after about seven to ten days, or at about 14 days,

and the binding of the at least one cell stimulatory polypeptide to the cell surface results in stimulation of the cell,

and results in the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell being more receptive to transduction by the lentiviral vector.

2. (previously presented): The method of claim 1 further comprising continuous contacting of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell *in vitro* or *ex vivo* with the lentiviral vector after the simultaneous contacting of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cells with the lentiviral vector and the at least one cell surface protein or receptor binding stimulatory polypeptide.

3. (previously presented): The method of claim 1 further comprising continuous contacting of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell *in vitro* or *ex vivo* with the at least one cell surface protein or receptor binding stimulatory polypeptide after the simultaneous contacting of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cells with the lentiviral vector and the at least one cell surface protein or receptor binding stimulatory polypeptide.

4. (canceled)

5. (previously presented): The method of claim 1 where the contacting with a lentiviral vector occurs more than once.

6. (previously presented): The method of claim 1 wherein the lentiviral vector is derived from a human immunodeficiency virus (HIV).

7. (canceled)

8. (previously presented): The method of claim 1 wherein the lentiviral vector comprises at least one cis-acting nucleotide sequence derived from the nef, gag, pol, env, vif, vpr, vpu, tat or rev genes.

9. (previously presented): The method of claim 8 wherein the cis-acting nucleotide sequence is not expressed or is a fragment or a mutant of the nef, gag, pol, env, vif, vpr, vpu, tat or rev genes.

10. (previously presented): The method of claim 1 wherein the lentiviral vector is derived from HIV-1 or HIV-2.

11. (previously presented): The method of claim 1 wherein the lentiviral vector is a pseudotyped vector.

12. (previously presented): The method of claim 11 wherein the pseudotyped vector comprises the vesicular stomatitis virus G envelope protein.

13. (previously presented): The method of claim 1 wherein the lentiviral vector is a chimeric vector comprising HIV sequences, wherein optionally the HIV sequences comprise HIV-1 and HIV-2 sequences.

14. (previously presented): The method of claim 1 wherein the primary lymphoid cell, myeloid cell or hematopoietic progenitor is (a) a CD3 positive cell; (b) a primary T cell or (c) a CD4 positive primary T cell.

15. (withdrawn): The method of claim 1 wherein the primary lymphoid cell, myeloid cell or hematopoietic progenitor is a monocyte or a CD14 positive cell.

16. (canceled)

17. (withdrawn): The method of claim 1 wherein said primary lymphoid cell, myeloid cell or hematopoietic progenitor cell is a CD34 positive cell or a CD34 positive hematopoietic precursor thereof.

18. (previously presented): The method of claim 14 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises or is a cell surface receptor binding polypeptide comprising an antibody that has the same cell surface binding specificity as a CD3 ligand; a CD28 ligand; a CD25 ligand; a CD71 ligand; a CD69 ligand; a CD49 ligand; or a cell surface protein or receptor binding stimulatory polypeptide having the same cell surface binding specificity as interleukin-2 (IL-2) or phytohemagglutinin (PHA).

19. (withdrawn): The method of claim 1 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises or is: a FLT-3 ligand, a thrombopoietin (TPO), a ligand for a TPO receptor, stem cell factor (SCF), a Kit ligand; or, an antibody or a polypeptide having the same cell surface binding specificity as FLT-3 ligand, thrombopoietin (TPO) ligand, stem cell factor (SCF) or Kit ligand.

20. (withdrawn): The method of claim 15 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises or is a monocyte surface receptor binding polypeptide comprising a granulocyte-macrophage colony-stimulating factor (GM-CSF), interleukin-4 (IL-4), tumor necrosis factor (TNF) alpha, interferon alpha or interferon gamma.

21. (withdrawn): The method of claim 1 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises CD34, CD3, CD14, CD28 or an antibody or other binding stimulatory polypeptide that has the same cell surface binding specificity as CD34, CD3, CD14, or CD28.

22. (previously presented): The method of claim 18, wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises a CD3 binding antibody or cell surface binding fragments thereof, a CD28 binding antibody or cell surface binding fragments thereof, combinations of the antibodies or cell surface binding fragments thereof, or polypeptides having the same cell surface binding specificities as the antibodies.

23. (withdrawn): The method of claim 22 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide is a T cell surface receptor binding polypeptide comprising (a) a combination of CD3 and CD28 antibodies immobilized on a bead or a surface, or (b) the antibody combination of (a), wherein the bead or surface comprises coated beads.

24-28. (canceled)

29. (previously presented): The method of claim 1 wherein the contacting the surface of the cells at the same time *in vitro* or *ex vivo* with both the lentiviral vector and the at least one cell surface protein or receptor binding stimulatory polypeptide further comprises (a) contacting the cell surface with a lentiviral vector for about 24 hours; or, (b) step (a) is repeated at least once.

30. (previously presented): The method of claim 1 wherein the lentiviral vector is present at an MOI of less than 500, or, the cells are transduced with the vector at a multiplicity of infection (MOI) such that the copies of vector per transduced cell is from about 1 to about 100.

31-32. (canceled)

33. (previously presented): The method of claim 1 wherein the contacting occurs *ex vivo*.

34. (currently amended): A method for stable transduction of a primary lymphoid cell, a myeloid cell or a hematopoietic progenitor cell ~~and/or~~ comprising:

- (a) isolating a primary lymphoid cell, a myeloid cell or a hematopoietic progenitor cell; and
- (b) contacting the cell of step (a) simultaneously *in vitro* or *ex vivo* with a lentiviral vector and [[an]]at least one cell stimulatory polypeptide that physically interacts with a receptor on the surface of the cell of step (a), wherein the at least one cell stimulatory polypeptide comprises or is an antibody, an antigen binding fragment, or a ligand;

wherein greater than ~~about 75%-90%~~ of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cells are stably transduced after about seven to ten days, or at about 14 days,

and the binding of the at least one cell stimulatory polypeptide to the cell surface results in stimulation of the cell

and results in the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell being more receptive to transduction by the lentiviral vector.

35. (previously presented): The method of claim 34 further comprising continuous contacting of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell *in vitro* or *ex vivo* with the lentiviral vector after the simultaneous contacting of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cells with the lentiviral vector and the at least one cell surface protein or receptor binding stimulatory polypeptide.

36-37. (canceled)

38. (previously presented): The method of claim 34 wherein the contacting with a lentiviral vector occurs more than once.

39. (canceled)

40. (previously presented): The method of claim 34 wherein the cell surface protein or receptor binding stimulatory polypeptide comprises an antibody, an antigen binding fragment, or a ligand.

41. (previously presented): The method of claim 34 wherein the lentiviral vector comprises at least one cis-acting nucleotide sequence derived from the nef, gag, pol, env, vif, vpr, vpu, tat or rev genes.

42. (previously presented): The method of claim 41, wherein the cis-acting nucleotide sequence is not expressed or is a fragment or a mutant of the nef, gag, pol, env, vif, vpr, vpu, tat or rev genes.

43. (previously presented): The method of claim 34 wherein the lentiviral vector is an HIV-derived vector.

44. (canceled)

45. (previously presented): The method of claim 34 wherein the lentiviral vector is pseudotyped with the vesicular stomatitis virus G envelope protein.

46. (canceled)

47. (previously presented): The method of claim 34 wherein the primary lymphoid cell, myeloid cell or hematopoietic progenitor is (a) a CD3 positive cell; (b) a primary T cell or (c) a CD4 positive primary T cell.

48. (withdrawn): The method of claim 34 wherein the primary lymphoid cell, myeloid cell or hematopoietic progenitor is a monocyte or is a CD14 positive cell.

49. (canceled)

50. (withdrawn): The method of claim 34 wherein said primary lymphoid cell, myeloid cell or hematopoietic progenitor cell is a CD34 positive cell or a CD34 positive hematopoietic precursor thereof.

51. (canceled)

52. (previously presented): The method of claim 34 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises or is a T cell surface receptor binding polypeptide comprising an antibody that has the same cell surface binding specificity as a CD3 ligand; a CD28 ligand; a CD25 ligand; a CD71 ligand; a CD69 ligand; a CD49 ligand; or a cell surface protein or receptor binding stimulatory polypeptide having the same cell surface binding specificity as interleukin-2 (IL-2) or phytohemagglutinin (PHA).

53. (withdrawn): The method of claim 34 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises or is: a FLT-3 ligand, a thrombopoietin (TPO), a ligand for a TPO receptor, stem cell factor (SCF), a Kit ligand; or, an antibody or a polypeptide having the same cell surface binding specificity as FLT-3 ligand, thrombopoietin (TPO) ligand, stem cell factor (SCF) or Kit ligand.

54-55. (canceled)

56. (previously presented): The method of claim 47 wherein the at least one cell surface protein or receptor binding stimulatory polypeptide comprises or is a T cell surface binding polypeptide comprising (a) CD3 antibodies or cell surface binding fragments thereof, CD28 antibodies and cell surface binding fragments thereof, combinations of the antibodies and cell

surface binding fragments thereof, or binding polypeptides that have the same cell surface binding specificities as the antibodies, or

(b) the cell surface protein or receptor binding stimulatory polypeptide of (a), wherein the least one cell surface binding stimulatory polypeptide comprises at least two of the cell surface binding polypeptides immobilized on a bead or a surface.

57. (withdrawn): The method of claim 56 wherein the at least one cell surface binding stimulatory polypeptide comprises a combination of CD3 and CD28 antibodies immobilized on coated beads.

58. (previously presented): The method of claim 34 further comprising culturing the cells under conditions conducive to growth and/or proliferation.

59. (currently amended): The method of claim 58 wherein the conditions comprise further incubation with a cell surface binding stimulatory polypeptide or a cytokine.

60. (canceled)

61. (previously presented): The method of claim 58 wherein the culturing is for about seven days.

62. (previously presented): The method of claim 58 wherein the culturing is for about 14 days.

63. (previously presented): The method of claim 34 wherein the contacting the cells with a lentiviral vector is for about 24 hours and is optionally repeated at least once.

64. (previously presented): The method of claim 34 wherein the lentiviral vector is present at an MOI of less than 500, or, the cells are transduced with the vector at a multiplicity of infection (MOI) such that the copies of vector per transduced cell is from about 1 to about 100.

65. (canceled)

66. (previously presented): The method of claim 34 wherein the contacting occurs *ex vivo*.

67. (previously presented): The method of claim 34 wherein the lentiviral vector is derived from a human immunodeficiency virus (HIV), wherein optionally the HIV is HIV-1 or HIV-2.

68. (previously presented): The method of claim 34 wherein the lentiviral vector is a chimeric vector comprising HIV-1 and HIV-2 sequences.

69. (currently amended): The method of claim 1 or claim 34, wherein greater than ~~80%, 85%, 89%, 90%~~, 91%, 92%, 93%, 94% or 95% of the cells are stably transduced after about 14 days.

70. (currently amended): The method of claim 34 wherein the cells in step (a) are isolated from an individual ~~is~~-infected with (a) a human immunodeficiency virus (HIV), or (b) HIV-1 or HIV-2.

71. (previously presented): The method of claim 70, wherein (a) the cells isolated from the HIV-infected individual are pre-stimulated with the at least one cell surface binding stimulatory polypeptide, or (b) the method of step (a) wherein the cells are pre-stimulated with the at least one cell surface binding stimulatory polypeptide within twenty four (24) hours prior to simultaneously contacting the cells *in vitro* or *ex vivo* with the lentiviral vector and the at least one cell surface binding stimulatory polypeptide.

72-82. (canceled)

83. (previously presented): The method of claim 1 or claim 34, wherein at least 75% of the cells remain stably transduced after about 14 days.

84. (canceled)

85. (withdrawn – currently amended): ~~The A method of claim 1 or claim 34, further comprising introducing the transduced cell genetic material into a living subject comprising~~ introduction of a cell transduced by the method of claim 1 or claim 34.

86. (withdrawn – currently amended): ~~The A method of claim 1 or claim 34, further comprising introducing the transduced cell genetic material into a tissue or an organ comprising introduction of a cell transduced by the method of claim 1 or claim 34.~~

87. (withdrawn – currently amended): ~~The A method of claim 1 or claim 34, further comprising introducing the transduced cell genetic material into a blastocyst comprising introduction of a cell transduced by the method of claim 1 or claim 34.~~

88. (currently amended): A method for stable transduction of a primary lymphoid cell, a myeloid cell or a hematopoietic progenitor cell with a lentiviral vector comprising contacting the cell surface of the primary lymphoid cell, myeloid cell or hematopoietic progenitor cell *in vitro* or *ex vivo* with a lentiviral vector and at least one cell surface polypeptide or receptor binding cell stimulatory polypeptide, wherein the lentiviral vector is pseudotyped, and wherein the at least one cell stimulatory polypeptide is an antibody, an antigen binding fragment, or a ligand, and

wherein after the contacting at least ~~about 75%-90%~~ of the cells are stably transduced after about seven to ten days, or at about 14 days, and optionally at least 75% of the cells remain stably transduced after about 14 days,

and the binding of the at least one polypeptide to the cell surface results in stimulation of the cell, and the binding of the cell surface receptor binding polypeptide to the cell surface receptor results in the cell being more receptive to transduction by the lentiviral vector.

89. (canceled)

90. (previously presented): The method of claim 88, wherein the lentiviral vector is pseudotyped with the Vesicular Stomatitis Virus envelope G (VSV-G) protein.

91-92. (canceled)

93. (withdrawn): The method of claim 1, claim 34, or claim 88, wherein the at least one cell surface binding stimulatory polypeptide comprises at least two cell surface binding polypeptides comprising an FLT-3 ligand; a thrombopoietin (TPO), a ligand for a TPO receptor; a Kit ligand; stem cell factor (SCF); interleukin-2 (IL-2); interleukin-4 (IL-4); granulocyte-macrophage colony-stimulating factor (GM-CSF); tumor necrosis factor (TNF) alpha; interferon alpha;

interferon gamma; or phytohemagglutinin (PHA); or antibodies that have the same cell surface binding specificity as FLT-3, thrombopoietin (TPO), or Kit ligand; a CD3 ligand; a CD28 ligand; a CD25 ligand; a CD71 ligand; a CD69 ligand; a CD49 ligand; or antibodies that have are the same cell surface binding specificity of CD3, CD25, CD28, CD49, CD69 or CD71 ligand.

94-96. (canceled)

97. (withdrawn): The method of claim 93, wherein the at least two cell surface binding stimulatory polypeptides comprise immobilized α CD3 and α CD28.

98. (previously presented): The method of claim 22, wherein the at least one cell surface binding stimulatory polypeptide comprises at least two cell surface binding polypeptides immobilized on a bead or a surface.

99. (previously presented): The method of claim 88, wherein the lentiviral vector is present at an MOI of less than 500, or, the cells are transduced with the vector at a multiplicity of infection (MOI) such that the copies of vector per transduced cell is from about 1 to about 100.

100. (withdrawn): A method for stable transduction of a CD3⁺ primary T cell comprising contacting the surface of the primary T cell at the same time *in vitro* or *ex vivo* with both a lentiviral vector and at least one polypeptide which binds the cell surface by binding to at least one cell surface receptor,

wherein at least about 75% of the cells are stably transduced after about seven to ten days, or at about 14 days,

and (a) if the primary T cell is CD4⁺ or CD8⁺, the at least one T cell surface receptor is a CD28 polypeptide,

and the binding of the primary cell surface receptor binding polypeptide to the cell surface receptor results in the cell being more receptive to transduction by the lentiviral vector.

101. (withdrawn): A method for stable transduction of a primary T cell with a lentiviral vector comprising

contacting the cell at the same time *in vitro* or *ex vivo* with a lentiviral vector and at least one cell surface receptor binding polypeptide, wherein the lentiviral vector is pseudotyped, and the pseudotyping comprises co-transfecting or co-infecting a packaging cell with both the lentiviral

vector genetic material and genetic material encoding at least one envelope protein of another virus or a cell surface receptor-binding polypeptide,

wherein (i) at least about 75% of the cells are stably transduced after about seven to ten days, (ii) at least about 75% of the cells are stably transduced at about 14 days, or (iii) at least 75% of the cells remain stably transduced after about 14 days,

and (a) if the primary T cell is $CD4^+$ or $CD8^+$, the at least one T cell surface receptor is a $CD28$ polypeptide,

and the binding of the cell surface receptor binding polypeptide to the cell surface receptor results in the cell being more receptive to transduction by the lentiviral vector.